REMARKS

This Amendment is in response to the Office Action, dated December 23, 2008 ("Office Action"). It is respectfully submitted that the application is in condition for allowance. Claims 1-42 are pending, with Claims 1-22 and 24-37 having been previously withdrawn. Claim 23 has been amended and claims 38-42 have been added by virtue of the present amendment. No new matter has been added. Allowance and reconsideration of the application in view of Applicants' amendment and the ensuing remarks are respectfully requested.

Claim 23 has been amended to specify that the mesenchymal stromal stem cells are "human." Support for this amendment may be found throughout the specification [*e.g.*, ¶ 131]. No new matter has been added.

Claim 23 was also amended to specify that the mesenchymal stromal stem cells are "encapsulated in a gel." Dependent claim 38 was also added to recite that the gel in which mesenchymal stromal cells are encapsulated may be an "alginate gel." Support for this amendment and the added dependent claim may be found throughout the specification [*e.g.*, ¶ 033]. No new matter has been added.

Claim 23 was also amended to recite that the mesenchymal stromal cells are exposed to "reduced oxygen tension." Dependent claims 39-42 were added to recite various ranges of oxygen tension. Support for this amendment and addition of the dependent claims may be found throughout the specification [*e.g.*, ¶ 122]. No new matter has been added.

In the Office Action, the Examiner acknowledged Applicants' election of the embodiment of the instant invention described in **Group V**, without traverse, (claim 23; drawn to a method of differentiating towards IVD cells by exposing cells to increasing pressures of up to 30psi) for prosecution on the merits. The Examiner also acknowledged Applicants' further election of the following species: (1) **bone marrow** in connection with claim 6; (2) **the application of load** (*i.e.*, step 8(c)) in connection with

claim 8; (3) **gels** in connection with claim 20; (4) **TGFβ** in connection with claim 28; and (5) **genes encoding inhibitors of cytokines** in connection with claim 15. The Examiner also acknowledged that Claim 23 reads on this species election and is generic with respect to each.

Claim 23 is rejected under 35 U.S.C. §112, first paragraph, as lacking enablement. The Examiner found Claim 23 to be directed to a method for causing mesenchymal stromal stem cells to differentiate towards IVD cells comprising exposing cultured mesenchymal stromal stem cells to increasing pressures of up to 30 psi. However, the Examiner asserts that undue experimentation would be required for one of skill in the art to make and/or use the claimed invention. In particular, the Examiner argues that Applicants' method has not provided any IVD differentiation marker on the MSSCs after the applied pressure alone. Furthermore, Examiner asserts that the specification fails to provide guidance, including parameters, for differentiation of MSSCs into IVD cells by exposing MSSCs to increasing pressure. The Examiner also contends that working examples are absent from the specification and the state of the art is unpredictable. Furthermore, the Examiner contends that the specification fails to provide any guidance for the production of proteoglycan at levels that would distinguish IVD cells from chondrocytes. Lastly, the Examiner argues that the breadth of the claim is considerable since it encompasses *any* animal species of IVD cells. Applicants respectfully traverse this rejection.

Applicants respectfully submit that the claims, as amended, comply with the enablement requirement under 35 U.S.C. §112, first paragraph. Furthermore, Applicants submit that the specification discloses the subject matter in such a way as to enable one skilled in the art to make and/or use the present invention. While Applicants do not acquiesce to the merits of the Examiner's rejection, in an effort to advance prosecution, claim 23 has been amended to: 1) specify that the mesenchymal stromal stem cells are "human;" 2) specify the mesenchymal stromal stem cells as being "encapsulated in a gel;" and 3) recite that the mesenchymal stromal cells are exposed to "reduced oxygen tension."

Applicants' proclaim that Examiner's concerns related to the ability of the

methods of the invention to induce cells to adopt an IVD phenotype in response to increasing pressure alone (the subject matter of the version of claim 23 under consideration in the Office Action) are addressed by the introduction of the limitations relating to the use of "cell encapsulation" and "reduced oxygen tension" for causing mesenchymal stromal stem sells to differentiate towards IVD cells. As noted by the Examiner, the specification clearly indicates "using more than one differentiation technique could improve differentiation" [¶ 126]. Since the amended claim 23 no longer refers to "differentiation of MSSCs into IVD cells by exposing the MSSCs into increasing pressure only" (see page 3, lines 19-20 of the Office Action), the Examiner should now acknowledge that the specification clearly discloses a working example of a method consistent with the amended claim 23. Accordingly, as amended, claim 23 now employs methods beyond the use of pressure only, incorporating the use of three different stimuli: pressure, gel encapsulation, and reduced oxygen tension, Applicants are of the opinion that in this respect the pending enablement rejection under §112, first paragraph, is now overcome.

Examiner's next objection states that "Applicants' method has not provided any IVD differentiation marker on the MSSCs after the applied pressure alone" (see page 4, lines 1-2 of the Office Action). In Example 4 of the present Application, two of the three differentiation techniques referred to in amended claim 23 are used (specifically, gel encapsulation and pressure loading of cells). As noted in the description of Example 4, this gives rise to cells with "phenotypes closely resembling natural NP cells" [¶ 126]. As set out in the specification [¶ 013], NP cells are an example of a group of cell types collectively referred to as IVD cells (in keeping with the language of claim 23). The specification provides ample guidance as to markers, the expression of which may be indicative of the generation of IVD cells. Merely by way of example, the production of IVD cells in accordance with the invention may be illustrated by expression of the proteoglycan versican [¶ 016]. Indeed, the Examiner refers to this marker on page 3, line 24 of the Office Action. The results of Example 4, shown in Figure 14, clearly illustrate that proteoglycan expression is approximately doubled in cells exposed to gel encapsulation and pressure loading differentiation techniques. Accordingly, the specification clearly illustrates that a technique utilizing only two of the three

differentiation techniques referred to in amended claim 23 is able to give rise to differentiation markers characteristic of IVD cells. Since the specification clearly indicates that the use of further differentiation techniques should be expected to provide an increasingly strong differentiation stimulus, the skilled artisan would appreciate that the disclosure of Example 4 is sufficient to provide a clear indication as to the effectiveness of the methods defined by the amended independent claim 23.

As amended, claim 23 no longer requires differentiation to be produced by "applied pressure alone," but instead uses a combination of specified differentiation stimuli. Applicants respectfully submit that substantive disclosure throughout the specification (*e.g.* Example 4) should be sufficient to overcome Examiner's objection in this respect.

Examiner has further objected to the claims, stating that "There is no evidence for the production of IVD differentiation markers at levels specific for IVD cells only" (see page 4, lines 23-24 of the Office Action) and that "At the time of the instant invention the art of MSSC cells differentiation to IVD cells by pressure alone was unpredictable" (see page 5, lines 6-8 of the Office Action). As stated prior, amended claim 23 does not rely upon pressure as the sole differentiation technique, but also utilizes gel encapsulation and reduced oxygen tension. Accordingly, Applicants assert that amendments made to claim 23 obviate the present objection. In parallel, Applicants would like to point out that the issue of markers characteristic of IVD cells, as opposed to chondrocytes, is not as unclear as Examiner indicates. Although Leung *et al.* may suggest that there was no single specific marker for NP cells, the present specification sets out a panel of markers that an artisan following the present invention may use to confirm an IVD cell phenotype. As taught by the specification, a matrix indicative of IVD cells may be characterized by the presence of one, or preferably more than one, of the markers in this panel [¶ 015] to [¶ 018].

Finally, Examiner has questioned the breadth of the claims embraced to any animal species of IVD cells, noting that undue experimentation would be required of one of skilled in the art to make and/or use such claims. Applicants assert that by amending claim 23 to specify that the mesenchymal stromal stem cells are limited to "human cells," Applicants obviate Examiner's rejection.

In light of the foregoing, all of the claims in the application are now believed to be allowable, and Applicants respectfully request withdrawal of this rejection under §112, first paragraph. Favorable consideration and a Notice of Allowance are earnestly solicited. If for any reason Examiner finds the application other than in condition for allowance, Examiner is requested to contact the undersigned attorney at the Los Angeles telephone number (213) 633-6800 to discuss the steps necessary for placing the application in condition for allowance.

Respectfully submitted,
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